

CHAPTER 7-6

WATER RELATIONS: REHYDRATION AND REPAIR



Figure 1. *Palustriella commutata* rehydrating in the spring runoff. Photo by Michael Lüth.

Uniqueness of Bryophytes

One striking difference between bryophytes and tracheophytes is that if you put a dry bryophyte into water, in most cases you will see an immediate change in turgor, and leaves will spread and take their normal hydrated position – one that presents the greatest surface area to the light and atmospheric CO₂. This is particularly striking in mosses from frequently dry habitats, such as *Hedwigia ciliata* from rocks or *Syntrichia ruralis* from sand. In many mosses, such as *Polytrichum* and *Syntrichia*, this ability to spread leaves when moist and appress them to the stem when dry is the result of enlarged or hyaline leaf base cells that absorb water easily and swell, forcing the leaf away from the stem.

Rehydration in mosses is generally very rapid, but some taxa are rather recalcitrant about getting wet inside. *Polytrichum piliferum* (Figure 2), common on sand in dry, exposed habitats, and *Schistidium apocarpum* (Figure 2), a rock-dweller, can require two hours to become saturated, while *Polytrichum juniperinum* (Figure 2), a soil moss with wider ecological amplitude than *P. piliferum*, can become saturated within three minutes (Larson 1981). Larson points out that the surface area to mass ratio is very important in determining the speed of rewetting (Figure 3). The cuticle seems to be another contributing factor in mosses like Polytrichaceae and Mniaceae.



Figure 2. **Top:** *Polytrichum piliferum* in hydrated state. Photo by Janice Glime. **Middle:** *Schistidium apocarpum* in its dry state with leaves wrapped around stem. Photo by Michael Lüth. **Bottom:** *Polytrichum juniperinum* in hydrated state. Photo by Janice Glime.

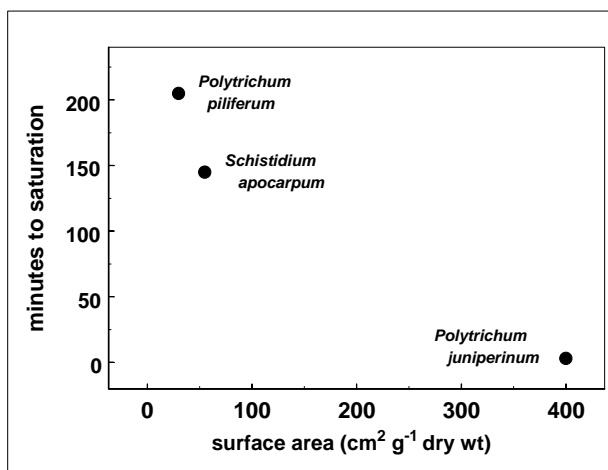


Figure 3. Relationship between surface area and time to saturation upon rewetting of three drought-tolerant mosses. Based on Larson (1981).

Resumption of Activity

Upon rehydration, desiccation-tolerant bryophytes generally resume normal activity quickly (Csintalan *et al.* 1999), whereas the resurrection plants among the tracheophytes in the same habitat take much longer (Peterson *et al.* 1994; Marschall & Proctor 1999). There are two general strategies that permit drought-tolerant plants to survive periods of desiccation: cellular protection and cellular repair. Those bryophytes that are tolerant of desiccation seem to succeed primarily because of their rapid cellular repair (Oliver *et al.* 1993). According to Oliver (1991), no novel **mRNAs** are recruited or favored for translation during desiccation. Rather, in *Syntrichia ruralis*, there is a loss of 25 **hydration** proteins (those present in a normal hydrated state), whereas 74 **rehydration** proteins are synthesized. This system, rather than protecting the moss from desiccation as in most tracheophytes, prepares bryophytes for repair. This is probably essential because their one-cell-thick leaves remain at full turgor, carrying out photosynthesis, then become desiccated very rapidly before going into a state of water stress and suspended metabolism (Proctor 2000b).

Antarctic mosses can suffer severe desiccation for prolonged periods. Rod Seppelt (Bryonet 2007) relates a story of an Antarctic *Grimmia*. A student had made a number of attempts at sectioning the dried moss without success. Seppelt suggested wetting the moss first and was amazed to discover, upon examination, that the cells were perfectly intact. When he re-examined the mosses that had been sitting on the lab bench for 15 months, but had been rewet for the sectioning, they had sprouted new shoots!

Breuil-Sée (1994) examined the cell interior upon rehydration of the thallose liverwort *Riccia macrocarpa* after 25 years of dehydration in a herbarium. Whereas most bryophytes revive to normal metabolism in a few hours, this 25-year-dry bryophyte required nine days. Cytological evidence of its revival included enlargement of **nucleoli**, evidence for protein synthesis. The dehydrated liverworts had few **mitochondria** and the chloroplasts lacked starch. Its preparation for desiccation was evidenced in granular cytoplasm with many **osmiophilic globules** (lipid-containing bodies in chloroplast), especially along the cell wall. Features already known for dry spores and seeds, such as presence of **plasmodesmata**, but absence of **dictyosomes** and **endoplasmic reticulum**, were evident. The transition of *R. macrocarpa* toward active metabolism upon rewetting was marked by 1) enlargement of nucleolus; 2) important modification of nucleus; 3) amplification of endoplasmic reticulum, Golgi, chloroplasts, mitochondria, and vacuoles; 4) disappearance of lipid reserves; 5) synthesis of starch in chloroplasts; 6) cytoplasm densification.

Protein Degradation

O'Mahony and Oliver (1999) compared the role of **ubiquitin** in the grass *Sporobolus stapfianus* and the desiccation-tolerant moss *Syntrichia ruralis* as a mediator of protein degradation. They found that in *S. stapfianus* the ubiquitin exhibited greater accumulation during drying and rehydration, but that it was hardly detectable in the desiccated tissue. A depletion of ubiquitin monomer levels indicates an increase in protein degradation. In *Syntrichia ruralis*, the transcripts were stable in the dried tissue. The

moss contrasted to the grass in that conjugated ubiquitin, indicative of proteins targeted for removal, was detectable in the moss only during slow drying, whereas it was present in all samples of the grass. O'Mahony and Oliver concluded that *S. ruralis* has stable ubiquitin transcripts that rapidly translate during rehydration to permit rapid initiation of cellular repair by degrading targeted proteins, whereas *S. stapfianus* requires several hours to replace its depleted ubiquitin supply.

Photosynthesis

Rehydration results in an initial period of rapid respiration (Dilks & Proctor 1976b). In several temperate/boreal bryophytes, this rapid period of respiration is followed by a progressive recovery of photosynthesis generally lasting 1-6 hours. *Anomodon viticulosus* reached its **compensation point** (photosynthesis = respiration) within a few minutes of hydration whereas it required about 4 hours for *Rhytidiadelphus loreus*. For desiccation-tolerant bryophytes such as *Anomodon viticulosus*, *Racomitrium lanuginosum*, and *Rhytidiadelphus loreus*, recovery of photosynthesis upon rehydration is rapid (Proctor & Smirnoff 2000). This rapid recovery necessarily requires pre-existing proteins; *de novo* protein synthesis is generally very limited (Proctor 2001).

Dhindsa (1985) determined that desiccation-tolerant mosses such as *Syntrichia ruralis* (Figure 4) remain active and fix CO₂ (dark fixation) at an undiminished rate until tissue losses are about 60% of the initial fresh mass, whereas in the intolerant *Cratoneuron filicinum* (Figure 5) dark fixation of CO₂ slowly declines as the moss dehydrates. After that, water stress occurs, the moss rapidly proceeds to suspended metabolism, and CO₂ fixation rapidly ceases. Following rehydration, *S. ruralis* immediately begins CO₂ fixation, but *C. filicinum* does not. For tracheophytes, this system has been perfected primarily in seeds that return from their suspended metabolism by metabolizing starches to sugars for the rapid supply of energy needed to grow and attain photosynthesis. Even in the desert ephemerals, the return process is slow and the frequency of wetting and drying suffered and survived by some desert bryophytes is unattainable by any tracheophyte (Proctor 2000b, 2001).



Figure 4. *Syntrichia ruralis* in hydrated state. Photo by Michael Lüth.



Figure 5. *Cratoneuron filicinum* in hydrated state. Photo by Michael Lüth.

Membrane Repair

Dry mosses are essentially inactive. Upon rehydration, the less tolerant bryophytes initially spend time in repairing membrane damage caused by the dehydration. This is exemplified by the period of 4 to 24 hours that elapse prior to normal photosynthesis and respiration (Peterson & Mayo 1975; Dilks & Proctor 1976b; Proctor 1981). But before that repair occurs, leakage of both photosynthate and mineral ions can be severe, especially during the first two minutes following addition of water (Bewley 1974; Gupta 1977). As in tracheophytes, the highly soluble K⁺ is readily leaked during desiccation (Minibayeva & Beckett 2001; Table 1), but in the bryophytes, much of it is retained by cation exchange sites on the cell walls (Bates 1997). Fortunately, these retained ions can be re-absorbed by the cells. Material leaked into a culture medium is taken back into the cell within one hour (Bewley & Krochko 1982). Furthermore, at least in some liverworts, some of the lost photosynthate is resorbed (Noailles 1978).

Table 1. Loss of K⁺ ions during rehydration following desiccation in bryophytes. **H** = hornwort; **LL** = leafy liverwort; **M** = moss; **TL** = thallose liverwort. Data from Minibayeva and Beckett (2001).

<i>Anthoceros natalensis</i> (H)	89%
<i>Pellia epiphylla</i> (TL)	83%
<i>Hookeria lucens</i> (M)	77%
<i>Dumortiera hirsuta</i> (TL)	55%
<i>Atrichum androgynum</i> (M)	45%
<i>Sphagnum auriculatum</i> (M)	38%
<i>Plagiochila natalensis</i> (LL)	21%
<i>Rhodobryum roseum</i> (M)	0%

In *Syntrichia ruralis*, slowly dried plants and undried controls lose only about half as much of electrolytes as rapidly dried plants (Bewley & Krochko 1982). However, *Cratoneuron filicinum* suffers more extensive loss under both slow and fast drying regimes and it is not reversible. Oliver and Bewley (1984b) interpreted these studies to mean that *Syntrichia ruralis* has membranes that undergo reversible changes during desiccation, but that these changes are incomplete when they are dried quickly. Upon rehydration it requires several minutes for the membranes to revert to their normal integrity. This mechanism apparently is not working in the desiccation-intolerant *Cratoneuron filicinum*.

Mechanical damage is probably the primary cause of desiccation damage in cells. Membranes necessarily become contorted and folded during drying and cell shrinkage. In *Syntrichia ruralis* pockets or **vesicles** (membranous sphere involved in transport or storage within cell) form on the **endoplasmic reticulum** complex system of membranous stacks involved in membrane production in cell). Oliver and Bewley (1984b) suggested that these vesicles provide membrane material to be used for immediate repair upon rehydration. Other features that can help protect a cell from mechanical damage during dehydration include small cell size, small or no vacuoles, lack of **plasmodesmata** (tiny, membrane-line channels between adjacent cells), flexible cell walls, and reduced osmotic pressure (Iljin 1953, 1957). However, there is not a strong correlation of these attributes with desiccation tolerant bryophytes. Bryophytes do have plasmodesmata, but electron microscopy is needed to discern them and few have been thus described; thus we cannot evaluate their correlation. Certainly some of the largest cells among bryophytes are those of the Hookeriaceae, a family of desiccation-sensitive mosses. And the Pottiaceae (including *Syntrichia ruralis*) generally have small cells and live in dry places. But the vacuole correlation also brings Iljin's suggested adaptations into question (Table 2), and even the cells of *Syntrichia ruralis* shrink but are too rigid to collapse when they dry. One problem in attempting to determine just what happens as the cells dry is that in order to "fix" them for examination, we must partially rehydrate the cells (Oliver & Bewley 1984b). Until another method is forthcoming, we cannot observe what a dry cell looks like.

Table 2. Relative cell and vacuole sizes among bryophytes listed by Oliver & Bewley (1984b).

	cell size	vacuoles
Desiccation tolerant		
<i>Ceratodon purpureus</i>	small	large
<i>Syntrichia ruralis</i>	small	small
<i>Neckera crispa</i>		small
<i>Pleurozium schreberi</i>	long & narrow	small
<i>Barbula torquata</i>	small	large
<i>Triquetrella papillata</i>	small	small
Desiccation sensitive		
<i>Cratoneuron filicinum</i>	long & narrow	small

The leakage problem causes bryophytes to be vulnerable during frequent wetting/drying events. During each rehydration event, the plant must repair its cell membranes, and that requires energy. Frequent events with insufficient recovery time will eventually exhaust the resources within the cells. Because much repair is needed upon rehydration, it is critical that dry mosses retain the ability to synthesize ATP upon rewetting (Krochko *et al.* 1979). In *Syntrichia ruralis*, normal levels of ATP are regained in as little as 30 minutes! On the other hand, the hydrophytic *Cratoneuron filicinum* slowly loses ATP after rewetting if the moss has been dried rapidly. Such behavior would prevent this moss from living in the desert, but poses no problem in its streamside habitat. However, Dhindsa (1985) suggested that it may be NADPH that is available immediately upon rehydration, produced by transhydrogenation from NADH during dark CO₂ fixation. Thus NADPH could be the important factor in repairing

cellular damage by reductive biosynthesis of membrane components and other cellular constituents.

When the membrane first begins repair, there is a period of enhanced respiration during which the cell organelles regain normal appearance (Noailles 1978). Membrane repair occurs during this period of enhanced respiration, stopping the leakage (Farrar & Smith 1976; Richardson & Nieboer 1980). This is possible because, unlike the case in tracheophytes, protein synthesis begins immediately (Dhindsa & Bewley 1978), undoubtedly because of the conservation of polyribosomes in desiccation-tolerant bryophytes. Nothing is known about the role of action potentials in bryophytes and their possible role in membrane repair (Bates 2000), although Trebacz *et al.* (1994) have shown that Ca²⁺ influx and Cl⁻ efflux in *Conocephalum conicum* result in depolarization of the cell membranes.

In an interesting contrast to this membrane repair scenario, Singh *et al.* (1984) concluded that membranes of *Syntrichia ruralis* remain intact during desiccation, at least down to 75% relative humidity (-400 bars). The cellular membranes retained their phospholipid bilayers, and during dehydration the cytoplasmic vesicles formed layers of membranes under the plasmalemma, appearing to fuse with the surface membrane. They concluded that the cellular membranes are conserved and ready to expand upon rehydration.

Thus, it is not surprising that Oliver *et al.* (1993) found that electrolyte leakage alone was not a reliable measure of desiccation tolerance in *Syntrichia ruralis*. Instead, Stewart and Lee (1972) reported that NADP-linked glyceraldehyde phosphate dehydrogenase is affected by desiccation, and Bewley and his coworkers (Bewley 1972, 1973a, b, 1974, 1979, Bewley & Gwozdz 1975) have carefully documented the loss of polyribosomes and their effect on the ability of the cells to synthesize proteins. Oliver *et al.* (1993) found that comparison of ability to synthesize protein in hydrated and desiccated-rehydrated mosses was the best measure of the capabilities of three *Syntrichia* species to repair damage and thus to exhibit tolerance to desiccation.

Even in such xerophytic taxa as *Syntrichia ruralis*, rapid drying causes visible injury, reduced total chlorophyll, reduction in chlorophyll *a:b* ratio, greatly enhanced electrolyte loss, and consequent inhibition of gross photosynthesis (Schonbeck & Bewley 1981a). Partial desiccation for 1-3 hours before rapid drying will eliminate this, suggesting that the moss requires time to prepare for its recovery. When *Syntrichia ruralis* and hydrophytic *Cratoneuron filicinum* have been dried rapidly, the chloroplasts and mitochondria swell and lose their integrity upon rewetting (Krochko *et al.* 1978, 1979), but *S. ruralis* regains normal appearance within 24 hours, whereas *C. filicinum* loses its cell contents and shows considerable cell degradation. However, if the cells are dried more slowly (*e.g.* 12 hours at 75% RH), both species recover within 24 hours. Dhindsa and Bewley (1978) attribute the ability of *Syntrichia ruralis* to survive this swelling of organelles to their ability to synthesize or retain sufficiently the enzymes needed for repair.

Deltoro *et al.* (1998a) compared recovery in seven desiccation-tolerant bryophytes (Figure 5; *Hedwigia ciliata*, *Hypnum cupressiforme*, *Leucodon sciuroides*, *Orthotrichum cupulatum*, *Pleurochaete squarrosa*, *Porella*

platyphylla, and *Syntrichia ruralis*) with that of seven desiccation-intolerant bryophytes (*Barbula ehrenbergii*, *Cinclidotus aquaticus*, *Conocephalum conicum*, *Lunularia cruciata*, *Palustriella commutata* (Figure 1), *Philonotis calcarea*, & *Rhynchostegium riparioides*). All seven desiccation-tolerant bryophytes experienced full recovery,

with many cellular activities back to normal rates within two hours (Deltoro *et al.* 1998a; Marschall & Proctor 1999). However, those species from the hydric and mesic habitats, the desiccation-intolerant ones, were unable to restore their photochemical activity.



Figure 6. Drought-tolerant bryophytes in the study by Deltoro *et al.* (1998a). **Left, top:** *Hedwigia ciliata*, **Left, Middle:** *Leucodon sciuroides*, **Left, bottom:** *Pleurochaete squarrosa*, **Right, top:** *Orthotrichum cupulatum*, **Right, middle:** *Hypnum cupressiforme*, **Right bottom:** *Porella platyphylla*. Photos by Michael Lüth.

Photodamage

For the most desiccation-tolerant mosses, those from **xeric** (dry) habitats, **fluorescence** (emission of light of longer wavelength due to absorbance of light from outside source) levels upon rehydration indicate that the photosynthetic apparatus is fully functional, unlike that of mosses from **hydric** (wet) and **mesic** (moderate) habitats (Deltoro *et al.* 1998a; Marschall & Proctor 1999). **Photoinhibition** (inhibition of photosynthesis by light) is a well-known consequence of desiccation because the **light**

quenching is greatly diminished or absent. Only the desiccation-tolerant bryophytes exhibited photo-quenching at low water content in these experiments. Deltoro and coworkers (1998a, b) suggest that this loss of photosynthetic capability in **mesophytic** bryophytes might be not only a consequence of photoinhibition, but also a result of membrane damage, as indicated by the large K^+ leakage. In desiccation-tolerant taxa, they suggest, the ability to enhance the dissipation of thermal energy during

dehydration might permit them to take advantage of the erratic water supply in places like the desert and decrease the problems of photodamage during the dehydration stage, thus permitting them to recover quickly.

Even drought-intolerant bryophytes may not die following total desiccation. My experience with boiling

Fontinalis and with dead-looking mosses following snow-melt is that seemingly dead bryophytes may have living cells that initiate new growth. The desiccated tissues may not recover, but a few cells may be all that are needed to continue the population.

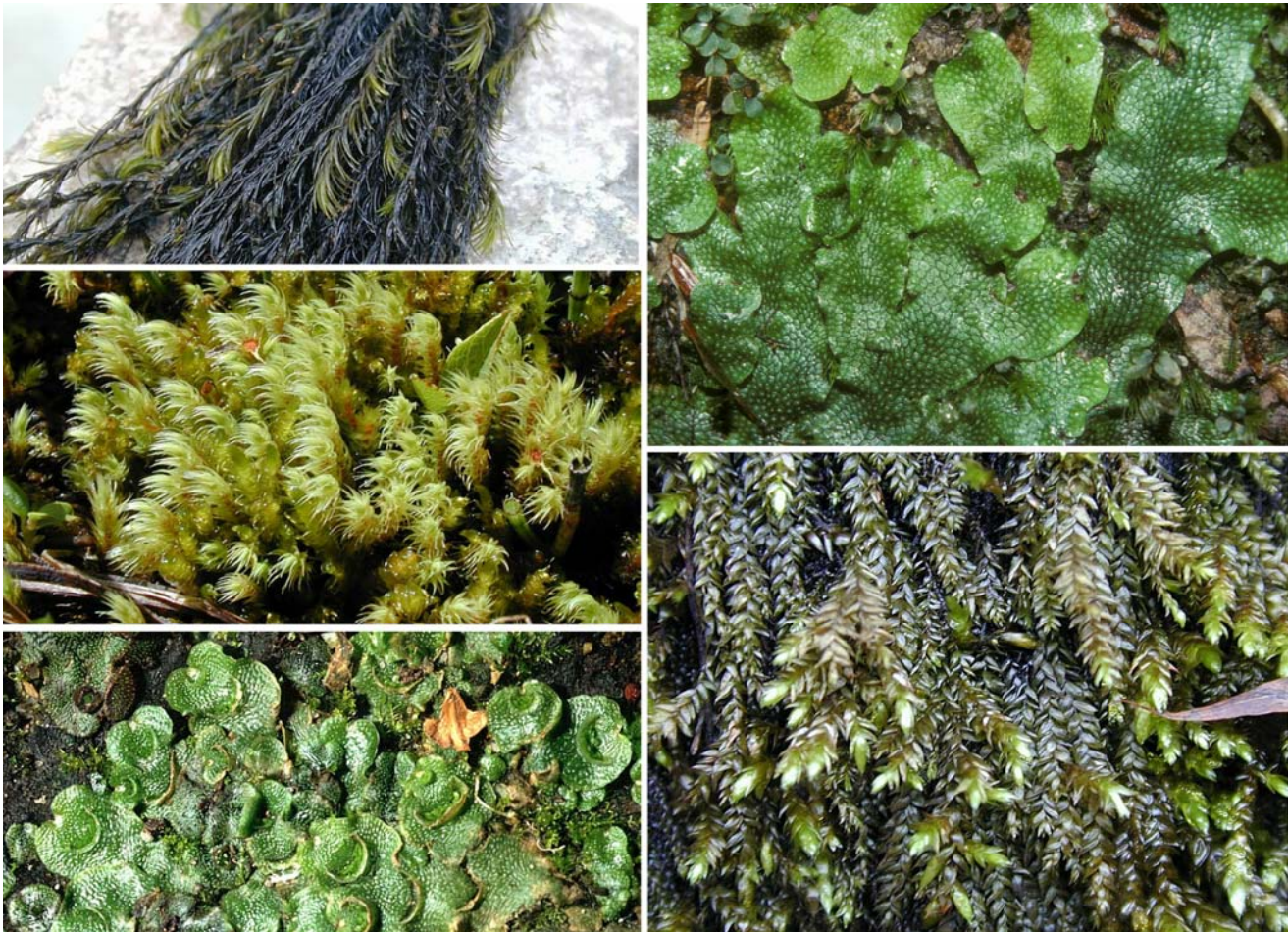


Figure 7. Desiccation-intolerant bryophytes in the study by Deltoro *et al.* (1998a). **Left, top:** *Cinclidotus aquaticus*, **Left, middle:** *Philonotis calcarea*, **Left, bottom:** *Lunularia cruciata*, **Right, top:** *Conocephalum conicum*, **Right, bottom:** *Rhynchostegium riparioides*. Photos by Michael Lüth; *Conocephalum conicum* photo by Janice Glime.

Architectural Changes

We know that many bryophytes, including *Syntrichia ruralis*, undergo multiple architectural changes as they dry (Hamerlynck *et al.* 2000). This results in changes to the surface reflectance. Hamerlynck *et al.* found a sigmoidal relationship between the relative humidity and the deviation of the moss mat temperature from its dew point, indicating a slow, then rapid, then slow change in the temperature of the mat, and a concomitant change in its water loss. The conditions of drying affect the ability of this species to use thermal dissipation of excess light energy, thus affecting potential damage to the chlorophyll.

Hamerlynck *et al.* (2002) later found that *Syntrichia ruralis* grown in high light intensity has greater desiccation tolerance than plants grown in the shade, but that those plants growing in the shade may benefit from their longer periods of metabolic activity and greater acquisition of resources, permitting them to adjust sufficiently to canopy openings and other disturbances.

Chloroplast Recovery

Wood and coworkers may have a partial answer to the recovery of the chloroplasts following desiccation (Wood & Oliver 1999; Wood *et al.* 1999; Zeng & Wood 2000; Zeng *et al.* 2002). As we have already discussed, there is a change in gene expression during rehydration of *Syntrichia ruralis*, suggesting that new proteins are being made. It appears that some of these proteins may account for the rapid chlorophyll recovery. We now understand that the moss prepares for its desiccation and rehydration events by altering gene expression in response to desiccation, then altering translational controls as it rehydrates. When the drying rate has been slow, **mRNPs** (messenger ribonucleoprotein particles) are formed in the drying plants, and within these particles they sequester **rehydrin** mRNA. It appears that one of these rehydrins may be responsible for the production of antioxidants during rehydration (Oliver *et al.* 1997). It is the production of these mRNPs that makes slow dehydration so important to the recovery

(Oliver 1996). If the moss is dried rapidly, it must make these when it rehydrates.

Wood and coworkers (1999) supported this discovery that *Syntrichia ruralis* has an active recovery mechanism that is induced by rehydration. It makes a set of polypeptides that are not present at any time except during rehydration. These polypeptides are products of a large number of as yet unidentified plant genes and 71% of these are unknown in other plant phyla.

Among these are most likely the cDNA *Rp115* identified by Zeng and Wood in 2000 and which is conserved as mRNA in desiccated gametophytes, and two additional cDNA units (*Elipa* & *Elipb*), both of which have significant similarity to Early Light-Inducible Proteins (ELIP; Zeng *et al.* 2002). The ELIP group (coded by *Elip* genes) includes over 100 stress-inducible proteins (Heddad & Adamska 2002). They are produced in response to light stress and accumulate in photosynthetic membranes where they have a photoprotective function. They are closely related to the light-harvesting chlorophyll *a/b*-binding antenna proteins of photosystems I and II. Because of the response of *Elipa* genes to slow desiccation, rapid desiccation/rehydration, salinity, ABA, and rehydration in high light, and the response of *Elipb* genes to ABA or rehydration in high light, Zeng *et al.* (2002) suggested that ELIPa and ELIPb provide an adaptive response to the photodamage that is likely to occur within a moss chloroplast during desiccation, most likely playing an important role in protecting and/or repairing the photosynthetic apparatus. In support of this hypothesis, Hutin and coworkers (2003) found that when they suppressed this rapid accumulation of ELIPs during high-light stress in a mutant of the flowering plant *Arabidopsis thaliana*, the leaves became bleached and cells suffered extensive photooxidative damage, but when the plant was permitted to accumulate ELIPs before the stress, they exhibited normal phototolerance. Hence, it appears that they do indeed perform a photoprotective function, either by binding the chlorophylls that are released during turnover of the pigment-binding proteins or by stabilizing the proper assembly of those proteins when they are being subjected to high-light stress.

When studying the grass *Sporobolus stapfianus*, Neale *et al.* (2000) found that *Elip* genes were expressed differently in tissues that were desiccation tolerant than in those that were desiccation sensitive and suggested that there are unique gene regulatory processes occurring as desiccation ensues, permitting different drought-responsive genes to be expressed at different stages during water loss. Since these genes have been identified in bryophytes, it is likely that Zeng *et al.* (2002) are correct in their suggestion of a photoprotective role during the dehydration state of bryophytes.

Frequency of Dehydration/Rehydration

Upon rehydration, it requires time to repair membranes and regain the energy lost. Oliver and Bewley (1984a) have demonstrated that in some mosses the first 24 hours are spent in repair, and it is only after that period that there is a net photosynthetic gain. For this reason, frequent short sequences of desiccation can be devastating to most species, whereas the same moss can endure long periods of desiccation. For example, *Barbula torquata* (Moore *et al.*

1982) recovered completely within one hour of rewetting after 18 months of desiccation at less than 5% relative water content. However, following short periods of desiccation, the integrity of the organelles was progressively lost, including membrane loss from chloroplasts and mitochondria. Repairing this damage resulted in delays in net photosynthetic gain.

Dilks and Proctor (1976b) promoted the understanding that frequency of desiccation can be more important than duration. Using 6 days wet, 1 day dry conditions compared to 1 day wet-6 days dry, 1 day wet-1 day dry, and 7 days wet-7 days dry for a period of 18 weeks, they showed that *Hylocomium splendens* grew equally well in continuous moist conditions and 6 days wet-1 day dry (32% relative humidity). However, there was little or no growth among the other treatments. In *Rhytidiadelphus loreus* (Figure 8), growth was best in continuously hydrated mosses, then 6 wet-1 dry day mosses, then 7 wet-7dry day mosses. There was essentially no growth in the other treatments. Responses by *Syntrichia ruralis* var. *arenicola* (syn. *Tortula ruraliformis*) were so variable that they could not be interpreted. However, they were able to conclude that 63 wet-dry cycles were not harmful and that constant moist conditions were in this highly desiccation-tolerant moss. *R. loreus*, unlike the other mosses, showed a **hardening** effect (process of increasing resistance to stress factor) *S. ruralis* is always drought-ready so hardening is not discernible), indicating less effect from drought as more droughts occurred.



Figure 8. *Rhytidiadelphus loreus*, a moss that undergoes drought hardening. Photo by Michael Lüth.

To test the impact of intermittent desiccation on xerophytic mosses, Mishler and Newton (1988) measured the success of germination of both fragments and spores of four *Tortula* species (*S. ruralis*, *S. princeps*, *S. norvegica*, *S. laevipila*) in continuous versus intermittent moisture. Only *S. princeps* fragments did slightly better under the intermittent moisture conditions, as did its spore germination. In all other species, the continuous hydration seemed beneficial to the spores. Establishment success was quite different. None of the spore-derived protonemata gave rise to stems (Mishler & Newton 1988). Fragments, however, produced numerous stems both from protonemata and directly from the fragments, independent of the hydration conditions. Most likely some other physiological or environmental cue was missing for the spore-derived protonemata.

We know that *Syntrichia ruralis* is capable of drought hardening (Schonbeck & Bewley 1981b). When subjected

to daily episodes of desiccation and rehydration, it develops a greater desiccation tolerance. However, the wet-dry cycle may be of less importance for boreal forest mosses. Hanslin and coworkers (2001) exposed *Dicranum majus* (Figure 9) and *Rhytidiadelphus loreus* to various watering regimes and found that responses, while differing greatly, lacked any consistent pattern. However, the relative growth rate increased with the length of the wet-dry cycle, provided the total number of wet and dry days remained equal, suggesting that these taxa probably would be unable to take advantage of night-time dew accompanied by daytime drought, but they are adapted to the more weekly or monthly wet-dry cycles typical of the boreal forest.



Figure 9. *Dicranum majus*. Photo by Michael Lüth.

Davey (1997) showed that Antarctic hydric mosses likewise are susceptible to damage by frequent wetting and drying, but that was not the case for the mesic and xeric mosses, which seemingly were adapted to frequent wet/dry cycles. All the mosses suffered a greater loss of photosynthetic rate as the duration of the dehydration periods increased. Davey suggested that mosses from the drier habitats were adapted to use short periods of rehydration. This is consistent with the use of late night/early morning moisture from clouds in xeric African montane sites and other habitats where nighttime dew is the major source of water. Csintalan and coworkers (2000) supported this concept with their work on *Syntrichia ruralis* in dry grasslands. They found that the moss absorbed progressive amounts of water through the night, permitting it to obtain about 1.5 hours of net photosynthetic gain immediately after dawn. Although this gain on many days may not be enough to offset the carbon loss during the remainder of the day, it does contribute to the overall carbon gain and may permit the moss to gain on a yearly scale when added to those occasions when more dew or moisture is available. In other species, high resistance is attained after several short exposures to drought (Clausen 1952; Abel 1956; Patterson 1964; Dilks & Proctor 1976a, b).

Implications

It appears that characteristics suggested for tracheophytes to permit them to survive desiccation (Iijin 1953, 1957) do not apply well to bryophytes. Rather, Oliver and Bewley (1984b) suggested that tolerant species must do three things to survive drying: (1) limit damage to a level that can be repaired; (2) maintain physiological integrity of the cell so metabolism can quickly reactivate during rehydration; (3) put repair mechanisms into effect

upon rehydration, especially to regain integrity of membranes.

Summary

Bryophyte gametophytes recover from desiccation by the actions of numerous **rehydration proteins**, including **rehydrins**, and **rapid membrane repair**. The rapidity is dependent upon slow dehydration that gives the bryophyte time to make mRNPs and is provided by a rehydration-inducible recovery mechanism in which new proteins are synthesized rapidly (Oliver 1996). The rapid recovery is complemented by enlargement of the nucleolus, amplification of the endoplasmic reticulum, Golgi, chloroplasts, mitochondria, and vacuoles, disappearance of lipid reserves, and synthesis of starch in chloroplasts during rewetting.

Short periods of rehydration between frequent drying periods deplete resources and are more harmful than long dry periods, issuing foreboding for moss gardeners.

Acknowledgments

This chapter has benefitted from the help of Beth Scafone and Medora Burke-Scoll, who helped me tow the line in explaining things without leaving too much to one's imagination, but at the same time not repeating myself.

Literature Cited

- Abel, W. O. 1956. Die Austrocknungsresistenz der Laubmoose. Anz. Osterr. Akad. Wiss. Math. Naturwiss. Kl. 165: 619-707. In: During, H. J. 1979. Life Strategies of Bryophytes: a preliminary review. *Lindbergia* 5: 2-18.
- Bates, J. W. 1997. Effects of intermittent desiccation on nutrient economy and growth of two ecologically contrasted mosses. *Ann. Bot.* 79: 299-309.
- Bates, J. W. 2000. Mineral nutrition, substratum ecology, and pollution. In: Shaw, A. J. and Goffinet, B. (eds.). *Bryophyte Biology*, Cambridge University Press, Cambridge, UK, pp. 248-311.
- Bewley, J. D. 1972. The conservation of polyribosomes in the moss *Tortula ruralis* during total desiccation. *J. Exper. Bot.* 23: 692-698.
- Bewley, J. D. 1973a. Desiccation and protein synthesis in *Tortula ruralis*. *Can. J. Bot.* 51: 203-206.
- Bewley, J. D. 1973b. The effects of liquid nitrogen temperatures on protein and RNA synthesis in the moss *Tortula ruralis*. *Plant Sci. Letters* 1: 303-308.
- Bewley, J. D. 1974. Protein synthesis and polyribosome stability upon desiccation of the aquatic moss *Hygrohypnum luridum*. *Can. J. Bot.* 52: 423-427.
- Bewley, J. D. 1979. Physiological aspects of desiccation tolerance. *Ann. Rev. Plant Physiol.* 30: 195-238.
- Bewley, J. D. and Gwozdz, E. A. 1975. Plant desiccation and protein synthesis II. On the relationship between endogenous adenosine triphosphate levels and protein-synthesizing capacity. *Plant Physiol.* 55: 1110-1114.
- Bewley, J. D. and Krochko, J. E. 1982. Desiccation-tolerance. In: Lange, O. L., Nobel, P. S., Osmond, C. B., and Ziegler, H. (eds.). *Physiological Plant Ecology. II. Water Relations and Carbon Assimilation*. Encyclopedia of Plant Physiology, New Series, vol. 12 B, Springer-Verlag, Berlin, Heidelberg, pp. 325-378.

- Breuil-Sée, A. 1994. Reviviscence d' un bryophyte en anhydrobiose depuis un quart de siècle: critères cytologiques d' aptitude à la reviviscence de thallus de *Riccia macrocarpa* Lev. [Reviviscence of a bryophyte in anhydrobiosis for a quarter of a century: cytological criteria of reviviscence ability in *Riccia macrocarpa* Lev. thalli.]. *Compt. Rend. Acad. Sci. (Paris) Ser. III Sci. Vie* 317: 245-252.
- Clausen, E. 1952. Hepatics and humidity, a study of the occurrence of hepatics in a Danish tract and the influence of relative humidity on their distribution. *Dansk Bot. Ark.* 15: 5-80.
- Csintalan, Z., Proctor, M. C. F., and Tuba, Z. 1999. Chlorophyll fluorescence during drying and rehydration in the mosses *Rhytidiadelphus loreus* (Hedw.) Warnst., *Anomodon viticulosus* (Hedw.) Hook. and Tayl. and *Grimmia pulvinata* (Hedw.) Sm. *Ann. Bot.* 84: 235-244.
- Csintalan, Z., Takács, Z., Proctor, M. C., Nagy, Z., and Tuba, Z. 2000. Early morning photosynthesis of the moss *Tortula ruralis* following summer dew fall in a Hungarian temperate dry sandy grassland. *Plant Ecol.* 151: 51-54.
- Davey, M. C. 1997. Effects of continuous and repeated dehydration on carbon fixation by bryophytes from the maritime Antarctic. *Oecologia* 110: 25-31.
- Deltoro, V. I., Calatayud, A., Gimeno, C., and Barreno, E. 1998a. Water relations, chlorophyll fluorescence, and membrane permeability during desiccation in bryophytes from xeric, mesic, and hydric environments. *Can. J. Bot.* 76: 1923-1929.
- Deltoro, V. I., Calatayud, A., Gimeno, C., Abadia, A., and Barreno, E. 1998b. Changes in chlorophyll a fluorescence, photosynthetic CO₂ assimilation and xanthophyll cycle interconversions during dehydration in desiccation-tolerant and intolerant liverworts. *Planta* 207: 224-228.
- Dhindsa, R. S. and Bewley, J. D. 1978. Messenger RNA is conserved during drying of the drought-tolerant moss *Tortula ruralis*. *Proc. Nat. Acad. Sci. USA* 75: 842-846.
- Dilks, T. J. K. and Proctor, M. C. F. 1976a. Seasonal variation in desiccation tolerance in some British bryophytes. *J. Bryol.* 9: 239-247.
- Dilks, T. J. K. and Proctor, M. C. F. 1976b. Effects of intermittent desiccation on bryophytes. *J. Bryol.* 9: 249-264.
- Dhindsa, R. S. 1985. Non-autotrophic CO₂ fixation and drought tolerance in mosses. *J. Exper. Bot.* 36: 980-988.
- Farrar, J. F. and Smith, D. C. 1976. Ecological physiology of the lichen *Hypogymnia physodes*. III. The importance of the rewetting phase. *New Phytol.* 77: 93-103.
- Gupta, R. K. 1977. A study of photosynthesis and leakage of solutes in relation to the desiccation effects in bryophytes. *Can. J. Bot.* 55: 1186-1194.
- Hamerlynck, E., Tuba, Z., Csintalan, Z., Nagy, Z., Henebry, G., and Goodin, D. 2000. Diurnal variation in photochemical dynamics and surface reflectance of the desiccation-tolerant moss, *Tortula ruralis*. *Plant Ecol.* 151: 55-63.
- Hamerlynck, E. I., Csintalan, Z., Nagy, Z., Tuba, Z., Goodin, D., and Henebry, G. I. 2002. Ecophysiological consequences of contrasting microenvironments on the desiccation tolerant moss *Tortula ruralis*. *Oecologia* 131: 498-505.
- Hanslin, H. M., Bakken, S., and Pedersen, B. 2001. The impact of watering regime and ambient relative humidity on the effect of density on growth in two boreal forest mosses, *Dicranum majus* and *Rhytidiadelphus loreus*. *J. Bryol.* 23: 43-54.
- Heddad, M. and Adamska, I. 2002. The evolution of light stress proteins in photosynthetic organisms. *Compar. Funct. Genomics* 3: 504-510.
- Hutin, C., Nussaume, L., Moise, N., Moya, I., Kloppstech, K., and Havaux, M. 2003. Early light-induced proteins protect *Arabidopsis* from photooxidative stress. *Proc. Natl. Acad. Sci.* 100: 4921-4926.
- Iljin, W. S. 1953. Causes of death of plants as a consequence of loss of water: Conservation of life in desiccated tissue. *Bull. Torrey Bot. Club* 80: 166-167.
- Iljin, W. S. 1957. Drought resistance in plants and physiological processes. *Ann. Rev. Plant Physiol.* 8: 257-274.
- Krochko, J. E., Bewley, J. D., and Pacey, J. 1978. The effects of rapid and very slow speeds of drying on the ultrastructure and metabolism of the desiccation-sensitive moss *Cratoneuron filicinum* (Hedw.) Spruce. *J. Exper. Bot.* 29: 905-917.
- Krochko, J. E., Winner, W. E., and Bewley, J. D. 1979. Respiration in relation to adenosine triphosphate content during desiccation and rehydration of a desiccation-tolerant and a desiccation-intolerant moss. *Plant Physiol.* 64: 13-17.
- Larson, D. W. 1981. Differential wetting in some lichens and mosses: The role of morphology. *Bryologist* 84: 1-15.
- Marschall, M. and Proctor, M. C. F. 1999. Desiccation tolerance and recovery of the leafy liverwort *Porella platyphylla* (L.) Pfeiff.: Chlorophyll-fluorescence measurements. *J. Bryol.* 21: 257-262.
- Minibayeva, F. and Beckett, R. P. 2001. High rates of extracellular superoxide production in bryophytes and lichens, and an oxidative burst in response to rehydration following desiccation. *New Phytol.* 152: 333-341.
- Mishler, B. D. and Newton, A. E. 1988. Influences of mature plants and desiccation on germination of spores and gametophytic fragments of *Tortula*. *J. Bryol.* 15: 327-342.
- Moore, C. J., Luff, S. E., and Hallam, N. D. 1982. Fine structure and physiology of the desiccation-tolerant mosses, *Barbula torquata* Tayl. and *Triquetrella papillata* (Hook. F. and Wils.) Broth., during desiccation and rehydration. *Bot. Gaz.* 143: 358-367.
- Neale, A. D., Blomstedt, C. K., Bronson, P., Le, T. N., Guthridge, K., Evans, J., Gaff, D. F., and Hamill, J. D. 2000. The isolation of genes from the resurrection grass *Sporobolus stapfianus* which are induced during severe drought stress. *Plant Cell Environ.* 23: 265-277.
- Noailles, M. 1978. Etude ultrastructurale de la récupération hydrique après une période de sécheresse chez une Hypnobryale: *Pleurozium schreberi* (Willd.) Mitt. *Ann. Sci. at. Ser.* 12: 19, 249-265.
- O'Mahony, P. J. and Oliver, M. J. 1999. The involvement of ubiquitin in vegetative desiccation tolerance. *Plant Molec. Biol.* 41: 657-667.
- Oliver, M. J. 1991. Influence of protoplasmic water loss on the control of protein synthesis in the desiccation-tolerant moss *Tortula ruralis*. *Plant Physiol.* 97: 1501-1511.
- Oliver, M. J. 1996. Desiccation tolerance in vegetative plant cells. *Physiol. Plant.* 97: 779-787.
- Oliver, M. J. and Bewley, J. D. 1984a. Plant desiccation and protein synthesis. IV. RNA synthesis, stability, and recruitment of RNA into protein synthesis during desiccation and rehydration of the desiccation-tolerant moss, *Tortula ruralis*. *Plant Physiol.* 74: 21-25.
- Oliver, M. J. and Bewley, J. D. 1984b. Desiccation and ultrastructure in bryophytes. *Advances in Bryology* 2: 91-131.
- Oliver, M. J., Mishler, B. D., and Quisenberry, J. E. 1993. Comparative measures of desiccation-tolerance in the *Tortula ruralis* complex. I. Variation in damage control and repair. *Amer. J. Bot.* 80: 127-136.

- Oliver, M. J., Wood, A. J., and O'Mahony, P. 1997. How some plants recover from vegetative desiccation: A repair based strategy. *Acta Physiol. Plant.* 19: 419-425.
- Patterson, P. M. 1964. Problems presented by bryophytism. *Bryologist* 67: 390-396.
- Peterson, G., Moyá, M. T., Zotz, G., Goslin, M., Kay, A., Maple, M., and Reich, A. 1994. Estimation of water loss rates in epiphytes. *Trop. Biol.: Ecol. Appr.* 94: 35-37.
- Peterson, W. L. and Mayo, J. M. 1975. Moisture stress and its effect on photosynthesis in *Dicranum polysetum*. *Can. J. Bot.* 53: 2897-2900.
- Proctor, M. C. F. 1981. Physiological ecology of bryophytes. *Adv. Bryol.* 1: 79-166.
- Proctor, M. C. F. 2000b. The bryophyte paradox: Tolerance of desiccation, evasion of drought. *Plant Ecol.* 151: 41-49.
- Proctor, M. C. F. 2001. Patterns of desiccation tolerance and recovery in bryophytes. *Plant Growth Reg.* 35(2): 147-156.
- Proctor, M. C. and Smirnoff, N. 2000. Rapid recovery of photosystems on rewetting desiccation-tolerant mosses: Chlorophyll fluorescence and inhibitor experiments. *J. Exper. Bot.* 51: 1695-1704.
- Richardson, D. H. S. and Nieboer, J. E. 1980. Surface binding and accumulation of metals in lichens. In: Cook, C. B., Pappas, P. W., and Rudolph, E. D. (eds.): *Cellular Interactions in Symbiotic and Parasitic Associations*. 5th Ann. Colloq. Coll. Biol. Sci., Ohio State Univ. Press, pp. 75-94.
- Schonbeck, M. W. and Bewley, J. D. 1981a. Responses of the moss *Tortula ruralis* to desiccation treatments. I. Effects of minimum water content and rates of dehydration and rehydration. *Can. J. Bot.* 59: 2698-2706.
- Schonbeck, M. W. and Bewley, J. D. 1981b. Responses of the moss *Tortula ruralis* to desiccation treatments. II. Variations in desiccation tolerance. *Can. J. Bot.* 59: 2707-2712.
- Singh, J., Blackwell, B., Miller, R., and Bewley, D. 1984. Membrane organization of the desiccation tolerant moss *Tortula ruralis* in several dehydrated states. Abstracts, Annual Meeting of the American Society of Plant Physiologists, 12-17 Aug., 1984. Univ. Calif. – Davis, p. 31.
- Stewart, G. R. and Lee, J. R. 1972. Desiccation injury in mosses. II. The effects of moisture stress on enzyme levels. *New Phytol.* 71: 461-466.
- Trebacz, K., Simonis, W., and Schönknecht, G. 1994. Cytoplasmic Ca^{2+} , K^+ , Cl^- and NO_3^- activities in the liverwort *Conocephalum conicum* L. at rest and during action potentials. *Plant Physiol.* 106: 1073-1084.
- Wood, A. J. and Oliver, M. J. 1999. Translational control in plant stress: The formation of messenger ribonucleoprotein particles (mRNPs) in response to desiccation of *Tortula ruralis* gametophytes. *Plant J.* 18: 359-370.
- Wood, A. J., Duff, R. J., and Oliver, M. J. 1999. Expressed sequence tags (ESTs) from desiccated *Tortula ruralis* identify a large number of novel plant genes. *Plant Cell Physiol.* 40: 361-368.
- Zeng, Q. and Wood, A. J. 2000. A cDNA encoding ribosomal protein RPL15 from the desiccation-tolerant bryophyte *Tortula ruralis*: mRNA transcripts are stably maintained in desiccated and rehydrated gametophytes. *Biosci. Biotechnol. Biochem.* 64: 2221-2224.
- Zeng, Q., Chen, X., and Wood, A. J. 2002. Two early light-inducible protein (ELIP) cDNAs from the resurrection plant *Tortula ruralis* are differentially expressed in response to desiccation, rehydration, salinity, and high light. *J. Exper. Bot.* 53: 1197-1205.